1P-32
Cyclosporin A suppresses up-regulated matrix metalloproteinase (MMP)-9 expression together with caspase-3/7 activity from keratinocyte in high calcium condition

Takashi Kobayashi*
Department of Dermatology, National Defense Medical college, Tokorozawa, Japan
*Contact author: kobat@ndmc.ac.jp

Keywords: gelatinase, keratinocyte, differentiation

Objective: Cyclosporin A is now administrated for not a few inflammatory conditions such as psoriasis and atopic dermatitis in dermatology. Among a variety of inflammatory conditions on which MMP-9 (gelatinase B) plays the role, we have focused on the apoptotic ones including abnormal keratinization in the epidermis, and the association between caspase-3/7 activity and gene regulatory mechanism of MMP-9 expression has been recently elucidated. This study presents the effect of cyclosporin A on the expressions of these enzymes from cultured keratinocyte in high calcium condition, which is considered to reflect hyperkeratois, high degree of epidermal differentiation, seen in many inflammatory skin diseases.

Methods: Human primary keratinocytes were cultured either in low or in high calcium concentration. In addition, cyclosporin A was added or not for each condition, and gelatinase activities together with caspase-3/7, -8, and -9 activities were analyzed.

Results: High calcium stimulation up-regulated both MMP-9 and caspase-3/7 expressions, which were suppressed by the addition of cyclosporine A in a dose-dependent manner.

Conclusions: These results indicate that the effect of cyclosporin A on inflammatory skin conditions is at least partly from the suppressions of MMP-9 and caspase-3/7 activities in hyperkeratosis.

REFERENCES

1P-33
IL-1 beta stimulates activin βA mRNA expression in human skin fibroblasts through MAP kinase pathways, NF-κB pathway and prostaglandin E2

KY Arai*, M Ono, C Kudo, Y Nomura. T Nishiyama
Scleroprotein Research Institute, Tokyo University of Agriculture and Technology, Fuchu, Tokyo 183-8509, Japan.
*Contact author: kojiarai@cc.tuat.ac.jp

Objectives: Accumulating evidences indicate that activin, a member of the transforming growth factor-β superfamily, plays important roles during skin wound healing. A noticeable increase in activin βA mRNA occurs in the injured skin and interleukin-1 (IL-1), which increases in the early phase of wound healing, is thought to be a probable factor inducing the increase in activin expression. The aim of this study is to reveal the mechanisms responsible for the IL-1β-induced activin βA mRNA expression in human skin fibroblasts.

Methods: Human skin fibroblasts were stimulated with IL-1β for 6 h, and levels of activin βA mRNA were examined by realtime PCR. To examine signaling pathways responsible for the IL-1β-induced activin expression, fibroblasts were pretreated with a JNK inhibitor SP600125, a p38 MAPK inhibitor SB202190, a MEK1/2 inhibitor U0126, an IKK2 inhibitor SC-514, a PKA inhibitor H-89, or a cyclooxygenase inhibitor indomethacin.

Results: Stimulation of fibroblasts with IL-1β considerably increased activin βA mRNA expression (more than 20-fold vs control at 1 ng/ml). SB202190, U0126 and SC-514 significantly suppressed the IL-1β-stimulated activin βA mRNA expression while SP600125 and H-89 failed to suppress it. Especially, SB202190 almost completely suppressed the IL-1β-stimulated activin βA mRNA expression. Because these effective inhibitors also suppressed IL-1β-stimulated expression of cyclooxygenase 2, a key enzyme for prostaglandin E2 (PGE2) synthesis, contribution of PGE2 to the IL-1β-stimulated activin βA mRNA expression was examined. Indomethacin significantly suppressed the IL-1β-stimulated activin βA mRNA expression to 40% of cells treated with IL-1β alone. Furthermore, stimulation of fibroblasts with 1 μM PGE2 for 6 h significantly increased activin βA mRNA.

Conclusion: The present study revealed that p38 MAP kinase, Erk1/2 and NF-κB pathways mediated the effect of IL-1β on activin βA mRNA expression. Furthermore, present results indicate that PGE2 is, at least in part, involved in the IL-1β-induced increase in activin expression.